Sample Application: Genetically Engineered Microorganisms

No CBI

U.S. DEPARTMENT OF AGRICULTURE
BIOTECHNOLOGY, BIOLOGICS, AND ENVIRONMENTAL PROTECTION

APPLICATION FOR PERMIT OR COURTESY PERMIT UNDER 7 CFR 340
(Genetically Engineered Organisms or Products)

INSTRUCTIONS: Complete this form and enclose the supporting materials listed on the reverse side. See page 3 for detailed instructions.

1. NAME AND ADDRESS OF APPLICANT
Dr. Jane Doe
Paige-Sullivan Boiotechnologies, Ltd.
6505 Belcrest Raod, Hyattsville, MD 20782
Area Code ( )

2. PERMIT REQUESTED ("X" one)
XXX☒ Limited - Interstate Movement
☐ Limited - Importation
☐ Release into the Environment
☐ Courtesy Permit

3. THIS REQUEST IS *("X" one)*
XXXX☒ New
☐ Renewal
☐ Supplemental

4. TELEPHONE NUMBER
(301) 436 - 7612

5. MEANS OF MOVEMENT
XXX☒ Mail ☐ Baggage or Handcarried
☐ Common Carrier By whom _____

6. GIVE THE FOLLOWING *(if applicable) (if more space is needed, attach additional sheet)*

	Scientific Name	Common Name	Trade Name	Other Designation
a. Donor Organism	Erwinia chrysanthemi CUPCPB 1237 (rif^r, $strp^r$)			
b. Recipient Organism	Escherichia coli HB 101			
c. Vector or Vector Agent	pBR322 and transformation			
d. Regulated Organism or Product	E. coli expressing pectate lyase (pCSR1)			
e. If product, list names of constituents.				

7. QUANTITY OF REGULATED ARTICLE TO BE INTRODUCED AND PROPOSED SCHEDULE AND NUMBER OF INTRODUCTIONS
one 2 ml culture vial

8. DATE *(or inclusive dates of period)* **OF IMPORTATION, INTERSTATE MOVEMENT, OR RELEASE**
Jan. 199X

9. COUNTRY OR POINT OF ORIGIN OF THE REGULATED ARTICLE
Dr. A. Collmer, Dept. of Plant Pathology
Cornell Univeristy, Ithaca, NY

10. PORT OF ARRIVAL, DESTINATION OF MOVEMENT, OR SPECIFIC LOCATION OF RELEASE
Hyattsville, MD

11. ANY BIOLOGICAL MATERIAL *(e.g., culture medium, or host material)* **ACCOMPANYING THE REGULATED ARTICLE DURING MOVEMENT**
culture media

12. APPLICANTS FOR A COURTESY PERMIT - STATE WHY YOU BELIEVE THE ORGANISM OR PRODUCT DOES NOT COME WITHIN THE DEFINITION OF A REGULATED ARTICLE

13. SEE REVERSE SIDE

I hereby certify that the information in this application and all attachments is complete and accurate to the best of my knowledge and belief.

False Statement: Falsification of any item on this application may result in a fine of not more than $10,000 or imprisonment for not more than 5 years or both. (18 U.S.C. 1001)

14. SIGNATURE OF RESPONSIBLE PERSON	15. PRINTED NAME AND TITLE	16. DATE
Jane Doe	Jane Doe, Regulatory Affairs Officer	10/29/9X

FOR APHIS USE ONLY

State Notification Sent	State Review Received	Permit Issued ☐ Yes ☐ No
Date of Determination	Permit No.	No. of Permit Labels Issued
Supplemental Conditions Enclosed ☐ Yes ☐ No		
Signature of BBEP Official	Date	Expiration Date

APHIS FORM 2000 (JUL 89) *Replaces PPQ Form 1001 which may be used.*

(continued on reverse)

	ENCLOSURES	ENCLOSED ("X")	IF PREVIOUSLY SUBMITTED, LIST DATE & PERMIT NO.
a.	Names, addresses, and telephone numbers of the persons who developed and/or supplied the regulated article		
b.	A description of the anticipated or actual expression of the altered genetic material in the regulated article and how that expression differs from the expression in the non-modified parental organism (e.g., morphological or structural characteristics, physiological activities and processes, number of copies of inserted genetic material and the physical state of this material inside the recipient organism (integrated or extrachromosomal), products and secretions, growth characteristics)		
c.	A detailed description of the molecular biology of the system (e g , donor-recipient-vector) which is or will be used to produce the regulated article	X	
d.	Country and locality where the donor organism, recipient organism, and vector or vector agent were collected, developed and produced.	X	
e.	A detailed description of the purpose for the introduction of the regulated article including a detailed description of the proposed experimental and/or production design.		
f.	A detailed description of the processes, procedures, and safeguards which have been used or will be used in the country of origin and in the United States to prevent contamination, release, and dissemination in the production of the. donor organism; recipient organism; vector or vector agent; constituent of each regulated article which is a product, and, regulated article.		
g.	A detailed description of the intended destination (including final and all intermediate destinations), uses, and/or distribution of the regulated article (e g., greenhouses, laboratory, or growth chamber location; field trial location, pilot project location, production, propagation, and manufacture location; proposed sale and distribution location).	X	
h.	A detailed description of the proposed procedures, processes, and safeguards which will be used to prevent escape and dissemination of the regulated article at each of the intended destinations.	X	
i.	A detailed description of the proposed method of final disposition of the regulated article	X	

APHIS FORM 2000 (Reverse)

13c. Total DNA was isolated from E. chrysanthemi, partially digested with Sau3A, sized on a sucrose gradient, and DNA fragments (4 kb) were pooled. pBR322 DNA was digested with BamH1 and dephosphorylated with alkaline phosphatase. Ligation of pBR322 and Sau3A-digested E. chrysanthemi DNA was with T4 DNA ligase. E. coli HB101 was transformed with recombinant plasmid DNA by CaCl2 (Mandel and Higa 1970). Transformants were screened for their ability to sink into pectate semisolid agar. Restriction mapping of the cloned DNA was performed by standard procedures.

13d. E. chrysanthemi was obtained from the Cornell University Collection of Phytopathogenic Bacteria. E. coli HB101 was obtained from ATCC and pBR322 was obtained from Bethesda Research Laboratories, Gaithersburg, MD.

13g. E. coli expressing pectate lyase will be manipulated in a laboratory setting. No experiments will be performed in growth chambers or greenhouses. The only experiments involving plant material will be the maceration of potato tuber discs as described in enclosed reprint (Collmer et al. 1985).

13h. All constructs were prepared according to the NIH Guidelines for Research Involving Recombinant DNA Molecules. All manipulations of recombinant bacteria were carried out in a laminar flow biosafety cabinet using "good microbiological practices." These experiments have been approved by our Institutional Biosafety Committee (IBC).

A copy of your IBC's approval of the research protocol for which this organism is being requested should accompany this application.

13i. All products containing the regulated article will be autoclaved prior to final disposal.

References

Mandel, M., Higa, A. 1970. Calcium dependent bacteriophage DNA infection. J. Mol. Biol. 53:159-162.

Collmer, A., Schoedel, C., Roeder, D. L., Reid, J. L., Rissler, J. F. 1985. Molecular cloning in Escherichia coli of Erwinia chrsyanthemi genes encoding multiple forms of pectate lyase. J. Bacteriol. 161:913-920.